



Short Report

Positive prostate-specific antigen (PSA) reaction in post-mortem rectal swabs: A cautionary note[☆]

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ABSTRACT

Prostate-specific antigen (PSA) tests are considered a valuable screening method for the forensic examination of semen in vaginal and rectal swabs of alleged victims of sexual abuse. Although these membrane tests have been also applied to autopsy specimens no study has assessed their reliability when performed on post-mortem (PM) rectal swabs from decomposed cadavers. The present study describes the results obtained with the Seratec® PSA Semiquant Kit test on 39 male and 10 female adult cadavers with no history of sexual assault and with a PM interval up to 136 days. Overall 64% of the 39 male cadavers tested positive for the PSA, the positive PSA reaction being more frequent in the 20 males with advanced decomposition than in the 19 males with no putrefaction signs (70% vs. 58%). The Phosphatesmo KM Paper Test® for detection of acid phosphatase (AP) gave a positive color reaction with 60% of the rectal swabs obtained from decomposed male cadavers. Both the PSA-test and the Phosphatesmo KM paper-test gave a negative result in each of the rectal samples from female cadavers. Y STR multiplex revealed no DNA other than that of the subject tested in the rectal swab positive for PSA. The results of the present study show that PSA membrane tests are unreliable and can be misleading when derived from male rectal samples obtained at autopsy.

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1. Introduction

Prostate-specific antigen (PSA) membrane tests are sensitive semi-quantitative immunoassay screening methods widely used in clinical settings for detection of seminal liquid in alleged victims of sexual abuse.^{1–4} Acid phosphatase (AP) paper test is another widely used detection test for seminal liquid.⁵ AP is an enzyme secreted by the prostate gland, but it is not unique to the prostate and can be found in other biological fluids.⁶

The PSA-test is recommended⁷ and has been used in autopsy cases involving suspected sexual crimes,^{8–10} but at the best of our knowledge, no study has so far assessed the reliability of this test when applied to rectal samples from decomposed cadavers obtained at autopsy.

The present study describes the results obtained with PSA membrane tests (Seratec®) and AP-tests (Phosphatesmo KM Paper®) from 39 males and 10 adult female cadavers with no history of sexual assault and with a post-mortem (PM) interval ranging from 2 to 136 days.

2. Materials and methods

2.1. Study material

Forty-nine subjects (39 males, 10 females) who underwent medico-legal autopsy at the Department of Forensic Medicine, University of Helsinki during 2004–2008 were selected for this study. The mean subject's age was in males 54 (SD: 15; range: 19–87 years) and in females 66 (SD: 14; range: 49–87 years). The subjects were classified in two groups: (a) 24 subjects (19 males, 5 females) with no visible signs of PM decomposition and (b) 25 subjects (20 males, 5 females) who displayed at external examination advanced putrefaction changes. The mean PM time for the subjects with advanced decomposition was 23 ± 25 SD (range: 8–136 days) and for the group with no putrefaction changes 5 ± 1 SD (range: 2–7 days).

2.2. Sampling and sample preparation

Three rectal swab samples were taken from the anal cavity of each cadaver and allowed to dry at room temperature. Extraction of the rectal swab content was performed in 1000 µl sterile Millipore® water for 30 min at room temperature. The extraction product was centrifuged at 10,000g for 1 min and 200 µl of the supernatant was added to the Seratec® PSA Semiquant Kit

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Table 1

Percentage of PSA membrane positive test (Seratec®) from PM rectal swabs in 49 subjects, by sex and PM changes.

Subjects N	Sex	Age (years)	PM interval (days)	PM ^b changes	Positive cases ^a	
					N	(%)
19	M	19–87	2–7	Fresh	11/19	58
20	M	32–79	8–136	Decomposed	14/20	70
5	F	49–80	3–5	Fresh	0/5	0
5	F	57–87	12–29	Decomposed	0/5	0

^a A case was defined positive when at least 2 of the 3 rectal swabs gave a strong positive reaction (>4 ng PSA/mL).^b Post-mortem.

according to the manufacturer's instructions.¹¹ A positive test result (>4 ng PSA/ml) is indicated by the formation of a red line in the test and control region of the membrane, the result being read 10 min after the addition of the supernatant. A few drops of the supernatant were added on a small piece of Phosphatesmo KM test paper (Phosphatesmo KM®, Macherey-Nagel GmbH & Co., Düren, Germany). A positive reaction gives a violet spot on a white background. All positive rectal samples were tested using the Y STR multiplex (PowerPlex®, Y System).¹²

2.3. DNA-extraction

Rectal swabs were rehydrated for 60 min in sterile Millipore® water and centrifuged for 3 min at 10,000g. The pellet was washed once in water and digested for 60 min at 56 °C in the presence of proteinase K (60 µg/0.2 ml final), DTT (4 mM final) and Chelex® resin (5% final). The tubes were then incubated at 80 °C for 8 min, centrifuged for 3 min and the supernatant was purified and concentrated in a QIAquick® (Qiagen) column before amplification.

3. Results and discussion

Overall 64% of the male cadavers tested positive for the PSA. Among the 19 male cadavers with no signs of putrefaction 58% tested positive compared with 70% in the 20 male cadavers with advanced decomposition changes. The membrane test did not detect PSA in the rectal samples from both decomposed and fresh female cadavers (Table 1).

Because the AP-test is used as a routine screening test for identification of semen in many laboratories, all swabs were also analyzed by this test. Twelve of the PSA positive swabs obtained from decomposed male cadavers showed a positive acid phosphatase reaction, while only four of the fresh male cadavers tested positive for AP (results not shown). A lower sensitivity and specificity of the AP-test compared with the PSA-test may explain this result. All swabs taken from female cadavers were negative both for PSA and AP.

In badly decomposed cadavers, autolysis of the prostate and PM changes of the rectum followed by the diffusion of the immunoreactive PSA and the AP enzyme through the rectal wall may explain the present results. On the other hand, it was surprising to find that more than half of the male cadavers with no advanced decomposition changes had also positive PSA reactivity in their anal swabs. Initially believed to be a prostate-specific protein, PSA is now known to be present in traces also in other tissues, including normal anal glands of males and urethral glands of both sexes.^{13,14} Release of PSA from normal epithelial cells of male anal glands following initial autolytic changes of the mucosa might account for the positive findings also during the early PM interval. No significant association was found between the positive cases and the subject's age. Using the Y STR multiplex (PowerPlex® System), no DNA other than that of the subject tested was detected in the rectal swabs positive for PSA.

4. Conclusion

PSA semi-quantitative membrane tests are considered a sensitive screening method for detection of seminal liquid in alleged male and female victims of sexual abuse in a clinical context. Conversely, the present study demonstrated that the results of PSA tests with a sensitivity comparable to the Seratec® PSA Semiquant Kit are unreliable and can be misleading when obtained at autopsy from male rectal samples, regardless of postmortem interval and the putrefaction stage. Also the AP Paper Test gave a positive color reaction with 60% of the rectal swabs obtained from decomposed male cadavers. Conversely, based on the results of this study, forensic biologists can extract material from postmortem female rectal swabs and should be confident that a PSA positive result stands for the presence of semen. However, a larger number of female cases need to be examined in order to substantiate this conclusion.

Conflict of Interest

We declare that we do not have any conflict of interests with regard to the submitted manuscript.

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Ethical Approval

None declared.

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